

PCT\_EP\_2003\_011551\_Sequence Listing.ST25.txt  
SEQUENCE LISTING

<110> Sloning Biotechnology GmbH

<120> Method for the manufacture of nucleic acid molecules

<130> S 10010 PCT

<140> EP 02023385.4

<141> 2002-10-18

<160> 61

<170> PatentIn version 3.1

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<221> misc\_feature

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acctgcnnnn nnnn

14

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<222> (7)..(11)

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<222> (7)..(16)

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<222> (7)..(17)

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<400> 24

ggcggannnn nnnnnnn

17

<210> 25

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<222> (7)..(17)

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<400> 25

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17

<210> 26

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<212> DNA

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<222> (8)..(15)

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<400> 26

cacctgcnnn nnnnn

15

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<223> splinker oligonucleotide in Fig. 1A and Fig. 3A

<400> 27

gtacgagacg cgcttttgcg cgtctcg

27

<210> 28

<211> 33

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> 1. anchor oligonucleotide in Fig. 1A and Fig. 3A

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<221> misc\_feature

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<222> (19)..(19)

<223> biotinylated nucleotide

<400> 28  
taccgccgaa gaggcgtttt cgcctcttcg gcg

33

<210> 29

<211> 60

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

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<221> misc\_feature

<223> sequence appears in Fig. 1B, Fig. 1C, Fig. 1D and Fig. 3B

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<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<220>

<221> misc\_feature

<222> (29)..(29)

<223> biotinylated nucleotide

<400> 29  
gcgcgtctcg taccgccgaa gaggcgtttt cgcctcttcg gcggtacgag acgcgctttt

60

<210> 30

<211> 33

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> left sequence in Fig. 1E

<400> 30  
gcggtacgag acgcgctttt gcgcgtctcg tac

33

<210> 31

<211> 27

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> right sequence in Fig. 1E, Fig. 3C and Fig. 3E

<220>

<221> misc\_feature

<222> (16)..(16)

<223> biotinylated nucleotide

<400> 31  
cgccgaagag gcgttttcgc ctcttcg

27

<210> 32

<211> 33

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> left sequence in Fig. 1F and Fig. 3E

<220>

<221> misc\_feature

<222> (19)..(19)

<223> biotinylated nucleotide

<400> 32  
cgctatcgaa gaggcgtttt cgcctcttcg ata

33

<210> 33

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

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<220>

<221> misc\_feature

<223> splinker oligonucleotide in Fig. 2A and Fig. 4A

<400> 33  
cgagacgcgc ttttgcgcgt ctcgt

25

<210> 34

<211> 41

<212> DNA

<213> Artificial Sequence



<220>

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<221> misc\_feature

<223> 1. anchor nucleotide in Fig. 2A and Fig. 4A

<220>

<221> misc\_feature

<222> (21)..(21)

<223> biotinylated nucleotide

<400> 34  
ccgtcatacg gatacgcggtt ttcgcgtatc cgtatgacgg a

41

<210> 35

<211> 66

<212> DNA

<213> Artificial Sequence

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<220>

<221> misc\_feature

<223> sequence appears in Fig. 2B, Fig. 2C, Fig. 2D and Fig. 4B

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<221> misc\_feature

<222> (32)..(32)

<223> biotinylated nucleotide

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<221> misc\_feature

<223> 5'-end and 3'-end are ligated

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gcgcgtctcg tccgtcatat ggatacgcgt tttcgcgtat ccgtatgacg gacgagacgc 60  
gctttt 66

<210> 36

<211> 33

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> left sequence in Fig. 2E, Fig. 2F, Fig. 4C, Fig. 4D and Fig. 4E

<400> 36  
cggacgagac gcgcttttgc gcgtctcgtc cgt 33

<210> 37

<211> 33

<212> DNA

<213> Artificial Sequence

<220>

<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> right sequence in Fig. 2E, Fig. 4C and Fig. 4D

<220>

<221> misc\_feature

<222> (17)..(17)

<223> biotinylated nucleotide

<400> 37  
catacggata cgcgttttcg cgtatccgta tga

33

<210> 38

<211> 41

<212> DNA

<213> Artificial Sequence

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<220>

<221> misc\_feature

<223> 2. anchor oligonucleotide in Fig. 2F and Fig. 4E

<220>

<221> misc\_feature

<222> (21)..(21)

<223> biotinylated nucleotide

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41

<210> 39

<211> 96

<212> DNA

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<221> misc\_feature

<223> sequence appears in Fig. 5A (left of text "Elongation product #1"

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<220>

<221> misc\_feature

<222> (47)..(47)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

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gcgcgctctcg tacgcgacgc gtcgtaagcc gtcccgaaga ggcgttttcg cctcttcggg 60  
acggcttacg acgcgtcgcg tacgagacgc gctttt 96

<210> 40

<211> 96

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

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<221> misc\_feature

<223> sequence appears in Fig. 5A (left of text "Elongation product #2"  
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<221> misc\_feature

<222> (47)..(47)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<400> 40  
gcgcgtctcg gtccggccta cgctagatcg atgccgaaga ggcgttttcg cctcttcggc 60  
atcgaactag cgtaggccgg accgagacgc gctttt 96

<210> 41

<211> 69

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> sequence appears in Fig. 5B ( left of text "Cut elongation product #1 with 3 nucleotide overhang at 5' end") and in Fig. 5C (left sequence left of text "Transition #1")

<400> 41  
ggacggctta cgacgcgtcg cgtagcagac gcgcttttgc gcgtctcgta cgcgacgcgt 60  
cgtaagccg 69

<210> 42

<211> 69

<212> DNA

<213> Artificial Sequence

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<221> misc\_feature

<223> sequence appears in Fig. 5B (left of text "cut elongation product #2 with 3 nucleotide overhang at 5' end") and in Fig. 5C (left sequence left of text "Transition #2")

<400> 42  
gcatcgaact agcgtaggcc ggaccgagac gcgcttttgc gcgtctcggt ccggcctacg 60  
ctagatcga 69

<210> 43

<211> 27

<212> DNA

<213> Artificial Sequence

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<220>

<221> misc\_feature

<223> sequence appears in Fig. 5C (right sequence left of text "Transition #1")

<220>

<221> misc\_feature

<222> (16)..(16)

<223> biotinylated nucleotide

<400> 43  
tcccgcgagacc gcgttttctgc ggtctcg 27

<210> 44

<211> 27

<212> DNA

<213> Artificial Sequence

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<221> misc\_feature

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<223> Sequence appears in Fig. 5C (right sequence left of text "Transition #2")

<220>

<221> misc\_feature

<222> (16)..(16)

<223> biotinylated nucleotide

<400> 44

tgccgagacc gcgttttcgc ggtctcg

27

<210> 45

<211> 96

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> sequence appears in Fig. 5D, Fig. 5E, Fig. 5F and Fig. 5G (in each case left of text "Elongation block #1")

<220>

<221> misc\_feature

<222> (47)..(47)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<400> 45

gcgcgtctcg tacgcgacgc gtcgtaagcc gtcccgagac cgcgttttcg cggtctcggg

60

acggcttacg acgcgtcgcg tacgagacgc gctttt

96

<210> 46

<211> 96

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> sequence appears in Fig. 5D, Fig. 5E, Fig. 5F, Fig. 7A (in each case left of text "Elongation block #2") and in Fig. 5H (right of text "Elongation block #2")

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<221> misc\_feature

<222> (47)..(47)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<400> 46

gcgcgtctcg gtccggccta cgctagatcg atgccgagac cgcgttttcg cggctctcggc 60

atcgaactag cgtaggccgg accgagacgc gctttt 96

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<220>

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<223> sequence appears in Fig. 5G (left of text "Eco31I cut Elongation block"), Fig. 5I (above text "Cut elongation block 1"), Fig. 7B and Fig. 7C (in each case left of text "Cut elongation block #1")

<400> 47  
ggacggccta cgacgcgtcg cgtacgagac gcgcttttgc gcgtctcgta cgcgacgcgt 60  
cgtaagcc 68

<210> 48

<211> 68

<212> DNA

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<221> misc\_feature

<223> sequence appears in Fig. 5H (right of text "Elongated transition anchor"), Fig. 5I (right of text "Cut elongation block 1"), Fig. 7B (left of text "Cut elongation block #2) and Fig. 7D (left of text cut elongation block #2")

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<221> misc\_feature

<222> (37)..(37)

<223> biotinylated nucleotide

<400> 48  
gtccggccta cgctagatcg atgccgagac cgcgttttgc cggctctcggc atcgaactag 60  
cgtaggcc 68

<210> 49

<211> 136

PCT\_EP\_2003\_011551\_Sequence Listing.ST25.txt

<212> DNA

<213> Artificial Sequence

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<221> misc\_feature

<223> sequence appears in Fig. 5J

<220>

<221> misc\_feature

<222> (67)..(67)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<400> 49	
gcgcgtctcg tacgcgacgc gtcgtaagcc gtccggccta cgctagatcg atgccgagac	60
cgcgttttcg cggctctggc atcgaactag cgtaggccgg acggcttacg acgcgtcgcg	120
tacgagacgc gctttt	136

<210> 50

<211> 106

<212> DNA

<213> Artificial Sequence

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<220>

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<223> sequence appears in Fig. 6A (left of text "Elongation produce #1"  
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<220>

<221> misc\_feature

<222> (52)..(52)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<400>	50		
gcgcgtctcg	tacgcgacgc	gtcgataagc	cgtctcatac ggatacgcgt tttcgcgtat 60
ccgtatgaga	cggcttatcg	acgcgtcgcg	tacgagacgc gctttt 106

<210> 51

<211> 106

<212> DNA

<213> Artificial Sequence

<220>

<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> sequence appears in Fig. 6A (left of text "Elongation product #2)

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<221> misc\_feature

<222> (52)..(52)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<400> 51  
 gcgcgtctcg gtccggccta cgctgagatc gatgccatac ggatacgcgt tttcgcgtat 60  
 ccgatatggca tcgaactcag cgtaggccgg accgagacgc gctttt 106

<210> 52

<211> 73

<212> DNA

<213> Artificial Sequence

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<220>

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<223> sequence appears in Fig. 6B (left of text "Cut elongation product #1 with 3 nucleotide overhang at 5' end") and Fig. 6C (left sequence left of text "Transition #1")

<400> 52  
 gacggcttat cgacgcgtcg cgtagcagac gcgcttttgc gcgtctcgta cgcgacgcgt 60  
 cgataagccg tct 73

<210> 53

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> sequence appears in Fig. 6C (left sequence left of text "Transition #1")

<220>

<221> misc\_feature

<222> (13)..(13)

<223> biotinylated nucleotide

<400> 53

cgagaccgcg ttttcgcggt ctcga

25

<210> 54

<211> 73

<212> DNA

<213> Artificial Sequence

<220>

<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> sequence appears in Fig. 6B (left of text "Cut elongation product #2 with 3 nucleotide overhang at 5' end") and in Fig. C (left of text "Transition #2")

<400> 54

catcgaactc agcgtaggcc ggaccgagac gcgcttttgc gcgtctcggt ccggcctacg

60

ctgagatcga tgc

73

<210> 55

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

PCT\_EP\_2003\_011551\_Sequence Listing.ST25.txt

<223> sequence appears in Fig. 6C (right sequence left of text "Transition #2")

<220>

<221> misc\_feature

<222> (13)..(13)

<223> biotinylated nucleotide

<400> 55  
cgagaccgcg ttttcgcggt ctcgg

25

<210> 56

<211> 98

<212> DNA

<213> Artificial Sequence

<220>

<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<223> sequence appears in Fig. 6D (left of text "Elongation block #1")

<220>

<221> misc\_feature

<222> (48)..(48)

<223> biotinylated nucleotide

<220>

<221> misc\_feature

<223> 5'-end and 3'-end are ligated

<400> 56  
gcgcgtctcg tacgcgacgc gtcgataagc cgtctcgaga ccgcgttttc gcggtctcga

60

gacggcttat cgacgcgtcg cgtacgagac gcgctttt

98

&lt;210&gt; 57

&lt;211&gt; 98

&lt;212&gt; DNA

&lt;213&gt; Artificial Sequence

&lt;220&gt;

&lt;223&gt; nucleic acid for the manufacture of nucleic acid molecules

&lt;220&gt;

&lt;221&gt; misc\_feature

&lt;223&gt; sequence appears in Fig. 6D (left of text "Elongation block #2")

&lt;220&gt;

&lt;221&gt; misc\_feature

&lt;222&gt; (48)..(48)

&lt;223&gt; biotinylated nucleotide

&lt;220&gt;

&lt;221&gt; misc\_feature

&lt;223&gt; 5'-end and 3'-end are ligated

&lt;400&gt; 57

gcgcgtctcg gtccggccta cgctgagatc gatgccgaga ccgcgttttc gcggtctcgg

60

catcgaactc agcgtaggcc ggaccgagac gcgctttt

98

&lt;210&gt; 58

&lt;211&gt; 96

&lt;212&gt; DNA

&lt;213&gt; Artificial Sequence

&lt;220&gt;

&lt;223&gt; nucleic acid for the manufacture of nucleic acid molecules

PCT\_EP\_2003\_011551\_Sequence Listing.ST25.txt

<220>

<221> misc\_feature

<223> sequence appears in Fig. 7A (left of text "Elongation block #1")

<220>

<221> misc\_feature

<222> (47)..(47)

<223> biotinylated nucleotide

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<223> 5'-end and 3'-end are ligated

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	cgcgacgcgt	cgtaagccgt cccgagccgg cgtttt 96

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<213> Artificial Sequence

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<222> (1)..(4)

<223> single-stranded overhang, not complemented by complementary strand

<220>

<221> misc\_feature

<222> (5)..(20)



<223> double-stranded nucleic acid, complemented by SEQ ID No. 48. The complementary strand continues in its 5'-direction with an overhang of 4 nucleotides (GCAT)

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ggacggccta cgacgcgtcg 20

<210> 60

<211> 20

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<213> Artificial Sequence

<220>

<223> nucleic acid for the manufacture of nucleic acid molecules

<220>

<221> misc\_feature

<222> (1)..(4)

<223> single-stranded overhang, not complemented by complementary strand

<220>

<221> misc\_feature

<222> (1)..(4)

<223> double-stranded nucleic acid, complemented by SEQ ID No. 47. The complementary strand continues in its 5'-direction with an overhang of 4 nucleotides (CAGG)

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<210> 61

<211> 108

<212> DNA

<213> Artificial Sequence

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<223> nucleic acid for the manufacture of nucleic acid molecules

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<221> misc\_feature

<223> sequence appears in Fig. 7D (right of text "Complementary overhang for subsequent transposition step")

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<222> (57)..(57)

<223> biotinylated nucleotide

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<223> 5'-end and 3'-end are ligated

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cggctctcggc atcgaactag cgtaggccgg acggccttacg acgcgtcg			108